

5 **SYSTEM FOR REAL TIME DETECTION OF NUCLEIC ACID**
 AMPLIFICATION PRODUCTS

10 **Abstract of the Invention**

 A system is provided for carrying out real time fluorescence-based measurements of nucleic acid amplification products. In a preferred embodiment of the invention, an excitation beam is focused into a reaction mixture through a surface, the
15 reaction mixture containing (i) a first fluorescent indicator capable of generating a first fluorescent signal whose intensity is proportional to the amount of an amplification product in the volume of the reaction mixture illuminated by the excitation beam and (ii) a second fluorescent indicator homogeneously distributed throughout the reaction mixture capable of generating a second fluorescent signal proportional to the volume of
20 reaction mixture illuminated by the excitation beam. Preferably, the excitation beam is focused into the reaction mixture by a lens through a portion of a wall of a closed reaction chamber containing the reaction mixture. The same lens is used to collect the first and second fluorescent signals generated by the first and second fluorescent indicators, respectively, in response to the excitation beam. The ratio of the fluorescent
25 intensities of the first and second fluorescent signals provides a stable quantitative indicator of the amount of amplification product synthesized in the course of the amplification reaction.

1